

ANSWER 1 OF 2 MEDLINE on STN DUPLICATE 1  
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 ACCESSION NUMBER: 1998197329 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 9536266  
 TITLE: Inhibition of HIV-1 replication by combined expression of gag dominant negative mutant and a human ribonuclease in a tightly controlled HIV-1 inducible vector.  
 AUTHOR: Cara A; Rybak S M; Newton D L; **Crowley R**; Rottschaefer S E; Reitz M S Jr; Gusella G L  
 CORPORATE SOURCE: Basic Research Laboratory, NCI, NIH, Bethesda, MD, USA.  
 SOURCE: Gene therapy, (1998 Jan) 5 (1) 65-75.  
 Journal code: 9421525. ISSN: 0969-7128.  
 PUB. COUNTRY: ENGLAND: United Kingdom  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals; AIDS  
 ENTRY MONTH: 199804  
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AB An HIV-1-based expression vector has been constructed that produces protective genes tightly regulated by HIV-1 Tat and Rev proteins. The vector contains either a single protective gene (HIV-1 gag dominant negative mutant (delta-gag)) or a combination of two different protective genes (delta-gag and eosinophil-derived neurotoxin (EDN), a human ribonuclease) which are expressed from a **dicistronic** mRNA. After stable transfection of CEM T cells and following challenge with HIV-1, viral production was completely inhibited in cells transduced with the vector producing both delta-gag and EDN and delayed in cells producing delta-gag alone. In addition, cotransfection of HeLa-Tat cells with an infectious HIV-1 molecular clone and either protective vector demonstrated that the HIV-1 packaging signals present in the constructs were functional and allowed the efficient assembly of the protective RNAs into HIV-1 virions, thus potentially transmitting protection to the HIV-1 target cells.

L25 ANSWER 2 OF 2 MEDLINE on STN DUPLICATE 2  
 ACCESSION NUMBER: 96211385 MEDLINE  
 DOCUMENT NUMBER: PubMed ID: 8649999  
 TITLE: High-level production of recombinant proteins in CHO cells using a **dicistronic** DHFR intron expression vector.  
 AUTHOR: Lucas B K; Giere L M; DeMarco R A; Shen A; Chisholm V; **Crowley C W**  
 CORPORATE SOURCE: Department of Molecular Biology, Genentech, Inc., South San Francisco, CA 94080-4990, USA.  
 SOURCE: Nucleic acids research, (1996 May 1) 24 (9) 1774-9.  
 Journal code: 0411011. ISSN: 0305-1048.  
 PUB. COUNTRY: ENGLAND: United Kingdom  
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)  
 LANGUAGE: English  
 FILE SEGMENT: Priority Journals  
 ENTRY MONTH: 199607  
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AB We have constructed expression vectors for Chinese hamster ovary (CHO) cells that produce both selectable marker and recombinant cDNA from a single primary transcript via differential splicing. These vectors produce stable CHO cell clones that, when pooled, produce abundant amounts of secreted recombinant proteins compared with the amounts produced by conventional expression approaches that have selectable marker and the cDNA of interest under control of separate transcription units. Our vectors divert most of the transcript to product expression while linking

it, at a fixed ratio, to dihydrofolate reductase (DHFR) expression to allow selection of stable transfectants. Pools of clones with increased expression of the product gene can be efficiently generated by selection in methotrexate. The high level of expression from pools allows convenient and rapid production of milligram amounts of recombinant proteins.

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